Data Evaluation Record on the Acute Toxicity of AE F159481 Technical (metabolite of Glufosinate Ammonium) to Fish, *Oncorhynchus mykiss*

EPA MRID Number 48444806

Data Requirement:	EPA DP Barcode	345709
	EPA MRID	48444806
	EPA Guideline	850.1075

Test material: **AE F159481 Technical (metabolite of Glufosinate Ammonium)** **Purity:** 98.9% w/w
Common name
Chemical name: IUPAC disodium methylphosphinato-acetate
CAS name
CAS No.
Synonyms

Primary Reviewer: Moncie Wright
Staff Scientist, Cambridge Environmental

Signature: Moncie V Wright
Date: 7/6/11

Secondary Reviewer: Teri S. Myers
Senior Scientist, Cambridge Environmental, Inc.

Signature: Teri S Myers
Date: 10/13/11

Primary Reviewer: Catherine Aubee
Biologist, US EPA/OPP/EFED/ERBIV

Signature: C Aubee
Date: 1 June 2012

EPA PC Code 128850

Date Evaluation Completed: 01-06-2012

CITATION: Heusel, R. 1997. Disodium methylphosphinato-acetate; substance, technical (metabolite of glufosinate ammonium) Code: AE F159481 00 1C99 0001 - Acute toxicity to rainbow trout (*Oncorhynchus mykiss*). Unpublished study performed and sponsored by Hoechst Schering AgrEvo GmbH, Frankfurt am Main, Germany. Laboratory Study Number: CE96/129. Study completed November 18, 1997.

EXECUTIVE SUMMARY:

In a 96-h acute toxicity study, rainbow trout, *Oncorhynchus mykiss*, were exposed to **AE F159481 Technical (metabolite of Glufosinate Ammonium)** at nominal concentrations of 0 (negative control), 9.9, 17.8, 31.7, 55.4, and 98.9 mg ai/L (corrected for purity) under static conditions. Mean-measured concentrations were <LOQ (<1.06, control), 10.5, 18.6, 33.8, 59.3, and 97.9 mg ai/L.

The 96-h LC_{50} was >97.9 mg ai/L. The NOAEC value, based on a lack of mortality and sub-lethal effects, was 97.9 mg ai/L.

Based on the results of this study, **AE F159481 Technical** would be classified as practically non-toxic to *Oncorhynchus mykiss* in accordance with the classification system of the U.S. EPA.

This toxicity study is scientifically sound and is classified as **acceptable**. It is consistent with the OCSPP Guideline 850.1075 requirements for an acute fish toxicity study with a glufosinate transformation product (MPA).

Results Synopsis (mean-measured)

Test Organism Size/Age(mean weight or length): 5 months; 1.7 g
Test Type (Flow-through, Static, Static Renewal): Static

LC_{50}: >97.9 mg ai/L 95% C.I.: N/A
NOAEC: 97.9 mg ai/L Probit Slope: N/A
EC_{50}: N/A

Statistical method: None used (reviewer's and study authors' results)

Endpoints calculated using mean-measured concentrations.

Endpoint(s) Affected: none

I. MATERIALS AND METHODS

GUIDELINE FOLLOWED: The test procedure followed recommended bioassay practices: OECD guideline no. 203: Fish, Acute Toxicity Test (adopted July 17, 1992); U.S. EPA Pesticides Assessment Guidelines, Subdivision E, §72-1: Acute toxicity test for freshwater fish; EPA Methods for Acute Toxicity Tests with Fish, Macroinvertebrates, and Amphibians (USEPA-660/3-75-009); and EU Directive 92/69/EWG Annex Part C: Methods for the determination of ecotoxicity; C.1. Acute toxicity to fish. The reviewer assessed the study methods and results according to U.S. EPA OPPTS 850.1075 and OECD No. 203, and similarities and/or deficiencies were described. One deficiency and three deviations from U.S. EPA OPPTS 850.1075 and/or OECD 203 were noted:

1. The study author did not analyze the water for particulate matter, pesticides, total organic carbon, metals, or chlorine content. OPPTS guidelines have established maximum concentrations for these parameters, and suggest that water quality be tested at least biannually. However, OECD guidelines do not suggest that these parameters be measured. Lack of water quality data can render a study invalid. However, no fish died or experienced sublethal effects.

2. The physico-chemical properties of the test material were not reported; OPPTS and OECD guidelines suggest that this information be provided by the study author.

3. The study author did not include a 15-30 minute transition period between light and dark conditions as recommended by OPPTS guidelines. OECD guidelines do not suggest a transition period.

4. Feed was only withheld for 24 hours before testing; OPPTS guidelines suggest that feed should be withdrawn 48 hours prior to testing. However, OECD guidelines only suggest a withdrawal period of 24 hours.

These deficiencies/deviations did not substantively impact the acceptability of this study.

COMPLIANCE: Signed and dated GLP, Quality Assurance and Data Confidentiality statements were provided. This study was conducted in compliance with the Principles of Good Laboratory Practice as adopted by OECD on 12 May 1981 [C(81)30 (Final)] and implemented at the national level.

A. MATERIALS:

1. Test material **AE F159481 Technical (metabolite of Glufosinate Ammonium)**

Description: White powder

Lot No./Batch No. : Not reported

Purity: 98.9% w/w

Stability of compound

under test conditions: Analytical verification of the test concentrations at time 0 yielded recoveries ranging from 102 to 109% of the nominal concentrations. At 96 hours (test termination), recoveries ranged from 96 to 109% of nominal. The test material was stable under the test conditions.

(OECD recommends water solubility, stability in water and light, pKa, Pow, and vapor pressure of test compound)

Storage conditions of test chemicals: Not reported.

Physicochemical properties of AE F159481 Technical (metabolite of Glufosinate Ammonium).

Parameter	Values	Comments
Water solubility at 20EC	Not reported	
Vapor pressure	Not reported	
UV absorption	Not reported	
pKa	Not reported	
Kow	Not reported	

2. Test organism:

Species: Rainbow trout, *Oncorhynchus mykiss* *EPA recommends a cold water species (preferably rainbow trout Oncorhynchus mykiss) and a warm water species (preferably bluegill sunfish Lepomis macrochirus). OECD recommends choice of species at discretion of testing laboratory.*

Age at test initiation: Juveniles (5 months old)

Weight at study initiation: 1.67 g *EPA recommends: mean 0.5 - 5 g.*

Length at study initiation: 4.8 cm (the longest fish was not twice as long as the shortest fish) *EPA recommends: Longest not > 2x shortest; OECD recommends 2.0 ∀1.0 cm for bluegill and 5.0 ∀1.0 cm for rainbow trout*

Source: Fish were obtained from the hatchery Oeseder Forellen, Sieben Quellen, Dr. Rosengarten, 49124 Oesede, Georgsmarienhutte, Germany, on July 10, 1996 and then maintained in the fish maintenance room of the laboratory/sponsor.
EPA recommends that all organisms be from the same source

B. STUDY DESIGN:

1. Experimental Conditions

a. Range-finding study A range-finding study was not reported.

b. Definitive Study

Table 1: Experimental Parameters

Parameter	Details	Remarks

		Criteria
Acclimation		
Period:	3 months; fish were reared under test conditions	*The recommended acclimation period is a minimum of 14 days; OECD guideline recommends a minimum of 12 days. Pretest mortality should be < 3% 48 h. prior to testing. OECD pretest mortality criteria: >10% = rejection of entire batch; ≥ 5 and ≤ 10% = continued acclimation for 7 days; <5% = acceptable.*
Conditions: (same as test or not)	Same as test (dilution water, temperature, pH, and dissolved oxygen)	
Feeding:	Fish were fed 6 times a week with a standard trout food (Kronen-Fish, Aminostart, Kirchhain, Germany), at a daily ration level of 2% of initial fish weight.	
Health: (any mortality observed)	The mortality in the stock culture 10 days prior to testing was 0.02%.	
Duration of the test	96 hours	
		The recommended test duration is 96 hours.
Test condition		
Static/flow-through	Static	*A reproducible supply of toxicant is recommended. Consistent flow rate is usually 5-10 vol/24 hours; meter systems should be calibrated before and after study and checked twice daily during test period.*
Type of dilution system - for flow-through method.	N/A	
Renewal rate for static renewal	N/A	
Aeration, if any	None during the test	
		Aeration is not recommended; OECD guideline recommends aeration. If aeration is necessary, test solutions must be analyzed periodically to verify exposure.

Parameter	Details	Remarks *Criteria*
Test vessel Material: (glass/stainless steel) Size: Fill volume:	 Stainless steel 50 L 50 L	 *Test vessel size is usually 19 L (5 gal) or 30 x 60 x 30 cm. Fill volume is usually 15-30 L of solution.*
Source of dilution water Quality:	The test water was a well aerated mixture of 50% filtered tap water and 50% deionized water. Both types of water were passed through sand and activated charcoal filters before being mixed together. The test water was aerated with compressed air to oxygen saturation and the temperature was adjusted to the test temperature.	*Recommended source of dilution water is soft, reconstituted water or water from a natural source. EPA does not recommend the use of dechlorinated tap water; however, its use may be supportable if the biological responses for the organisms and chemical analyses of residual chlorine meet conditions in the Agency=s 850.1010 guidelines for dilution water (http://www.epa.gov/opptsfrs/OPPTS_Harmonized/850_Ecological_Effects_Test_Guidelines/Draft/850.1010.pdf) Dilution water should be intensely aerated before the study. OECD permits dechlorinated tap water.*

Parameter	Details	Remarks *Criteria*
Water parameters: Hardness pH Dissolved oxygen Total Organic carbon Particulate Matter Metals Pesticides Chlorine Temperature {Salinity for marine or estuarine species} Intervals of water quality measurement	 1.53-1.56 mmol/L (154.4-156.1 mg/L as $CaCO_3$ 8.0-8.3 8.0-10.4 mg/L Not determined Not determined Not determined Not determined Not determined 12.7-13.8°C N/A Temperature, pH, and dissolved oxygen were measured daily. Additionally, the temperature of the control group was recorded continuously.	Water analysis for total hardness, acid binding capacity, and nitrite content were conducted on the first and last test day. Nitrite: <0.05 mg/L Conductivity: 355-436 µS/cm Acid binding capacity: 2.68-2.76 mmol HCl/L *Hardness:* *EPA recommends 40 - 48 mg/L as $CaCO_3$ (OECD recommends 10 - 250 mg/L)* *pH:* *EPA recommends 7.2 - 7.6; 8.0-8.3 for marine-stenohaline fishes, 7.7-8.0 for estuarine-euryhaline fishes, monthly range < 0.8); (OECD recommends pH 6.0 - 8.5)* *Dissolved Oxygen:* *EPA recommends: Static: ∃60% during first 48 hrs and ∃40% during second 48 hrs; flow-through: ∃60%; (OECD guideline recommends at least 80% saturation value).* *Temperature:* *EPA recommends 12 EC for coldwater species, 17 or 22 EC for warmwater species, and 22 ± 1 EC for estuarine/marine organisms. (OECD recommends 21 - 25°C for bluegill and 13 - 17°C for rainbow trout).* *Salinity:* *EPA recommends 30-34‰ (parts per thousand) for marine, 10-17‰ for estuarine fish, weekly range < 6‰.* *Water quality should be measured at beginning of test and every 48 hours.*
Number of replicates/groups: control: solvent control: treated ones:	 1 N/A 1	 *Recommended number of replicates include a control and five treatment levels. Each concentration should be 60% of the next highest concentration; concentrations should be in a geometric series.*
Number of organisms per replicate		

Parameter	Details	Remarks *Criteria*
/groups: control: solvent control: treated ones:	 10 N/A 10	*Number of organisms per replicate should be ∃10/concentration; OECD guideline recommends at least 7 fish/concentration.*
Biomass loading rate	0.33 g fish/L	*Recommended static conditions are #0.8 g/L at #17EC and #0.5 g/L at > 17EC. Recommended flow-through conditions are #1 g/L/day. OECD recommends a maximum of 1 g fish/L for static and semi-static, while higher rates are recommended for flow-through.*
Test concentrations: Nominal (uncorrected for purity): Nominal (corrected for purity): Mean-measured:	 0 (negative control), 10, 18, 32, 56, and 100 mg ai/L 0 (negative control), 9.9, 17.8, 31.7, 55.4, and 98.9 mg ai/L <LOQ (<1.06, control), 10.5, 18.6, 33.8, 59.3, and 97.9 mg ai/L	
Solvent (type, percentage, if used)	N/A	*The solvent should not exceed 0.5 ml/L for static tests or 0.1 ml/L for flow-through tests; OECD recommends that the solvent not exceed 100 mg/L.*
Lighting	16L:8D with no transition period reported Wide-spectrum fluorescent lighting	*The recommended photo period is 16 hours of light and 8 hours of dark with a 15-30 minute transition period. OECD recommends a photo period of 12 -16 hours.*
Feeding	Fish were not fed 24 hours prior to testing or during testing.	*Fish should not feed during the study.*
Recovery of chemical Frequency of determination Level of quantization	At time 0 and 96 hours, samples from the control and all treatment level solutions were analyzed via HPLC with UV detection (216 nm). Fortification samples, matrix blanks, and solvent blanks were analyzed concurrently. 1.06 mg/L	

Parameter	Details	Remarks *Criteria*
Level of detection	0.64 mg/L	
Positive control {if used, indicate the chemical and concentrations}	None used	
Other parameters, if any	None	

2. Observations:

Table 2: Observations

Parameter	Details	Remarks *Criteria*
Parameters measured including the sublethal effects/toxicity symptoms	- mortality - symptoms of intoxication	
Observation intervals	Every 24 hours	
		Observation intervals should be a minimum of every 24 hours.
Were raw data included?	Yes	
Other observations, if any	None	

II. RESULTS AND DISCUSSION:

A. MORTALITY:

There was no mortality in the control or any of the test levels during the 96 hours of the test.
EPA/OECD require pretreatment control mortality < 10%.. EPA requires that control or solvent mortality not exceed 10%. OECD requires that maximum-allowable control or solvent control mortality is 10% (or 1 mortality if 7 to 10 control fish are used) for a 96-h period of testing.

Table 3: Effect of AE F159481 Technical (metabolite of Glufosinate Ammonium) on Mortality of *Oncorhynchus mykiss*.

Treatment Mean-measured (and nominal) (mg ai/L)	No. of fish at start of study	Observation period					
		Day 1		Day 2		Day 4	
		No Dead	% mortality	No Dead	% mortality	No Dead	% mortality
Control (dilution water)	10	0	0	0	0	0	0
10.5 (9.9)	10	0	0	0	0	0	0
18.6 (17.8)	10	0	0	0	0	0	0
33.8 (31.7)	10	0	0	0	0	0	0
59.3 (55.4)	10	0	0	0	0	0	0
97.9 (98.9)	10	0	0	0	0	0	0
NOAEC	100 mg ai/L (based on nominal concentrations unadjusted for purity)						
LC_{50}	>100 mg ai/L (based on nominal concentrations unadjusted for purity)						
Positive control, if used mortality: LC_{50}:	N/A						

B. NON-LETHAL TOXICITY ENDPOINTS:

There were no sublethal effects in the control or any of the test levels.

Table 4: Sub-lethal Effect of AE F159481 Technical (metabolite of Glufosinate Ammonium) on *Oncorhynchus mykiss*.

Treatment Mean-measured (and nominal) (mg ai/L)	**Observation period**		
	Symptoms at Day 1	**Symptoms at Day 2**	**Symptoms at Day 4**
	% affected	**% affected**	**% affected**
Control (dilution water)	0	0	0
10.5 (9.9)	0	0	0
18.6 (17.8)	0	0	0
33.8 (31.7)	0	0	0
59.3 (55.4)	0	0	0
97.9 (98.9)	0	0	0
NOAEC	100 mg ai/L (based on nominal concentrations unadjusted for purity)		
LOAEC	Not determined		
EC_{50}	Not determined		
Positive control, if used % sublethal effect: EC_{50}:	N/A		

C. REPORTED STATISTICS:

There were no effects on mortality or sublethal effects, thus statistical analysis was not possible.

D. VERIFICATION OF STATISTICAL RESULTS:

Statistical Method: Due to a lack of mortality and sublethal effects, the reviewer visually determined toxicity values using the mean-measured test concentrations.

LC_{50}: >97.9 mg ai/L 95% C.I.: N/A

NOAEC: 97.9 mg ai/L

Probit Slope: N/A 95% C.I.: N/A

E. STUDY DEFICIENCIES:

The study author did not analyze the water for particulate matter, pesticides, total organic carbon, metals, or chlorine content.

F. REVIEWER'S COMMENTS:

The reviewer's results are in agreement with those of the study author's; there was no toxicity in this study. However, the reviewer used the mean-measured concentrations for the determination of toxicity values; therefore the

reviewer's results are presented in the Executive Summary and Conclusions sections of this DER.

The experimental start of the test was October 11, 1996, and the experimental termination date was October 16, 1996.

G. CONCLUSIONS:

This toxicity study is scientifically sound and is classified as **acceptable**. It is consistent with the OCSPP Guideline 850.1075 requirements for an acute fish toxicity study with a glufosinate transformation product (MPA). The 96-hour NOAEC and LC_{50} values were 97.9 and >97.9 mg ai/L (mean-measured), respectively.

III. REFERENCES:

U.S. Environmental Protection Agency (EPA), 1982. Pesticide Assessment Guidelines, Subdivision E, Hazard Evaluation: Wildlife and Aquatic Organisms. §72-1. Acute toxicity test for freshwater fish.

U.S. Environmental Protection Agency (EPA), 1975. Committee on Methods for Toxicity Tests with Aquatic Organisms. Methods for Acute Toxicity Tests with Fish, Macroinvertebrates, and Amphibians. EPA-660/3-75-009.

Organization for Economic Cooperation and Development. 1992. OECD Guideline for Testing of Chemicals. Guideline No. 203: Fish, Acute Toxicity Test. Adopted July 17, 1992.

EU Directive 92/69/EWG Annex Part C: Methods for the determination of ecotoxicity; C.1. Acute toxicity to fish.

U.S. Environmental Protection Agency (EPA), 1975. Brauhn, J.L., Schoettger, R.A. and Mueller, L.H. Acquisition and Culture of Research Fish: Rainbow Trout, Fathead Minnow, Channel Catfish and Bluegills. EPA-660/3-75-001.

Deutsches Institut fur Normung (DIN), 1989. Deutsche Einheitsverfahren zur Wasser-, Abwasser – und Schlammuntersuchung (German standard methods for the examination of water, wastewater and sludge). Normenausschub Wasserwesen (NAW) im DIN Deutshces Institut fur Normung e.V., Berlin, 1989.